

Three New Foliicolous Fungi on Sacred and Potent Ethno Medicinal Plant Indian Banyan from Shrawasti (U.P.) India

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ABSTRACT

This paper describes and illustrates *Alternaria banyan* sp. nov., *Asterina benghalensis* sp. nov., and *Drechslera fici* sp. nov. as new to Indian mycoflora from Shrawasti, (U.P.) India.

Keywords: Foliicolous Fungi, Indian Banyan, Sacred, Potent Ethno Medicinal Plant, Shrawasti, U.P.

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1. INTRODUCTION

The leaves provide a very suitable habitat for the growth and development of fungal pathogen by providing ample surface area and nutrient supply. Such leaf inhabiting fungi are known as Foliicolous Fungi and the invaded area of the leaf appears as leaf spot or leaf lesion. Taxonomic studies of such fungal forms have been generally considered as only of academic interest but the taxonomic treatment of a fungal organism is the first requirement for any studies concerning its biology. Correct identification of a fungus absolutely free from ambiguities is vital for its employment in applied disciplines. In fact without being equipped for ascertaining the correct identity of a fungal pathogen all studies concerning its phyto pathological aspects would be misleading. The weed and forest plants serve as reservoirs of leaf spot pathogens which on getting opportunity may spread to agriculture and horticulture plants.

India is located to the north of an equator, lies between 8° 4' and 37° 6' north latitude and 68° 7' and

97° 25' east latitude, measures 3214 kilometers from south to north and 2933 km from east to west, the total land area being 32,87,263 square kilometers. India is the one of the twelve mega biodiversity countries of the world lying in between Tropic of Capricorn and Tropic of Cancer, has two of the world's eighteen biodiversity hot spots located in the Western Ghats and in the Eastern Himalayas. The Himalayas rise as a virtual wall beyond the snow line, above the alluvial plain lies the Tarai strip, a seasonally marshy zone of sand and clay soil. The Tarai region has higher rain fall than the plains, and downward rushing rivers originating from the Himalayas slow down and spread out in the flatter tarai zone depositing fertile silt and reproductive means during the monsoon season and receding in the dry season. The Tarai, as a result has high water level and is characterized by moist subtropical conditions and a luxuriant turnover of green vegetation all the year around. The climatological and topographical conditions favor the luxuriant growth and development of foliicolous fungi. This North Tarai Region of U. P. is next only to

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Table 1

The comparative account *Alternaria tenuissima* and *Alternaria banyan* sp. nov.

| S. No. | Comparable Points | <i>Alternaria tenuissima</i> | <i>Alternaria banyan</i> sp. nov. |
|--------|-------------------|--|---|
| 1. | Infection spots | Hypogenous | Epigenous |
| 2. | Conidiophores | Arising singly or in group simple or branched mid pale brown, 1 to several conidial scars less geniculate, up to av 95 x 4-6 μ m. | Fasciculate, geniculate, unbranched monotretic rarely polytretic, thickened conidial scars, olive brown 40-110 x 4-7 μ m. |
| 3. | Conidia | Solitary or calenate obclavate or tapering gradually to the beak. Sometimes minutely verruculose slightly or not constricted at the septa, 22-95 x 8-9 20 μ m in diam. | Muriform, singly or in chains brown base obclavate to rounded, thick at broadest part constricted at the septa 25-70 x 14-20 μ m in diam. |

Eastern and Western Ghats, as one of the hottest spots for biodiversity in general and the diversity of fungal organism inhabiting plant in particular offers an ideal opportunity for the morphotaxonomic exploration of the fungal organism in general and foliicolous fungi in

particular. Keeping it in view the authors surveyed the North Tarai forests of Uttar Pradesh for Foliicolous Fungi on April 01, 2013 in locality of Gulra Rest House, Bhinga Forest Range representing Shrawasti Forest Division, Shrawasti.

2. SCOPE OF STUDY

The Foliicolous Fungi causes huge losses every year in different parts of the world. The fungal pathogens producing leaf spots infect a large variety of hosts including most of the crops, forests and other plants. The destruction caused by these enemies of leaves is a serious problem before us. The focus of this research is identification & documentation of these new foliicolous fungi will assist in the discovery of new fungicides and ideas to overcome from the severity of these enemies of nature as well as in the protection of floral diversity from the infection of these pathogens and also in the conservation of valuable flora of the area.

3. MATERIALS AND METHODS

During survey and collection, infected plant parts where noticed were collected carefully in the field and notes were made regarding their pathogen city, nature of colonies infection, locality, altitude. For each collection a separate field number was given. Each infected plant parts was collected separately in polythene bags along with host twig preferably with the reproductive parts to facilitate the identity of corresponding hosts. These collections were pressed neatly and dried in between blotting papers. The host plants were identified by matching them with authentic herbarium material and also consulting the experts.

In the laboratory, Hosagounder and Kapoor, 1984 nail polish technique was used to study the structural and morphological characters of fungi. Since the desired quality and quantity of nail polish is difficult to procure from the market, the problem is eased by preparing a xylene - thermocol

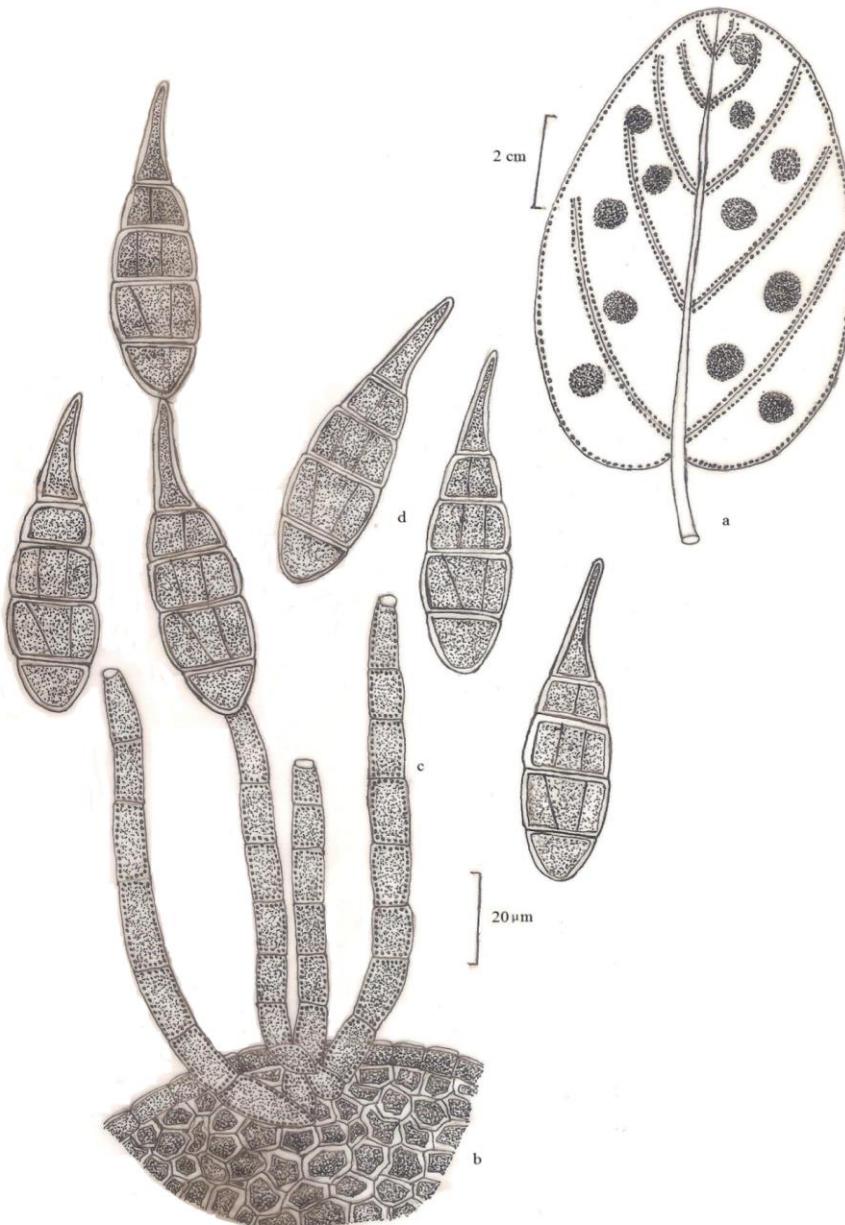


Figure 1

Alternaria banyan Mall and Kumar sp. nov.; a) Infected leaf, b) stroma, c) Conidiophores, d) Conidia

solution. Five ml or desired quantity of xylene is poured in a container, very bright and clean thermocol cut into minute pieces, added to xylene, mixed thoroughly till getting it to a particular consistency and poured it into air tight bottle for the use. A drop of xylene – thermocol solution applied on the selected colonies, carefully tinned the help of a fine brush without disrupting the colonies.

The treated colonies along with their host plants kept in dust free chamber for half an hour. When the applied solution dried, a thin colorless “film” or flip is formed with the colonies firmly embedded in it. The flip was lifted up with a slight pressure on the upper side of the leaves and just below the colonies on an edge of the flip eased and subsequently the entire flip peeled-off by using the thumb nail finger of the left hand. In case of hard host plants, the flip was eased off with the help of a razor or scalpel. A drop of DPX was added on a clean slide and flip was spread properly on it. Care was taken to avoid air bubbles while mounting. One or two more drops of DPX was again added on the flip and clean cover glass brings out the excess DPX and it was removed after drying. These slides were labeled and placed in dust free chamber for one or two days for drying.

Slides were prepared in cotton-blue lacto phenol mixture and were examined. Camera Lucida drawings were made and the morpho taxonomic determination of the taxa was done using available literature. The fungal taxa were identified using microscopic preparation. The fungal holotype specimen has been deposited for allotment of accession number from HCIO. The Mycobank No. from the Fungal Database Nomenclature and Species Banks has also been procured.

4. RESULT AND DISCUSSION

During our survey of the North Central Tarai Forests of Uttar Pradesh for foliicolous microfungi on April 01, 2013 in locality of Gulra Rest House we came across a plant *Ficus benghalensis* Linn., Indian Fig, the Indian Banyan, Bengal Fig, Bargad or Bar (Moraceae) considered Sacred and is called Vat Vriksha is also an important ethno medicinal plant. According to Ayurveda, it is astringent to bowels, useful in treatment of biliousness, ulcers, erysipelas, vomiting, vaginal complains, fever, inflammations, leprosy. According to Unani system of medicine, its latex is aphrodisiac, tonic, vulnerary, maturant, lessens inflammations, useful in piles, nose-diseases, gonorrhea etc. The aerial root is styptic, useful in syphilis, biliousness, dysentery, inflammation of liver etc. This plant is also useful in soil conservation. The leaves were found to be infected where dorsal surface of the leaves were found with dark brown to black thin infection spots coalescing to cover almost entire leaves with age.

On primarily stage of study three different types of spores were found in a single slide. On critical study the

leaves were found to be infected with plurivorous hyphomycetous fungi *Alternaria banyan* sp. nov., ascomycetous fungi *Asterina benghalensis* sp. nov. and a deuteromycetous fungi *Drechslera fici* sp. nov..

On critical examination and comparison with other known species *Asterina mysorensis* Hanf. And Thirum (1948) *Asterina* was to be found a new species. Hence it is described as *Asterina benghalensis* Mall and Kumar sp. nov.; *Alternaria* sp. has been reported on *Ficus benghalensis* from Khandala, Bombay. *Alternaria tenuissima* has been found to be reported on *Artocarpus heterophyllus* of the same family from Gorakhpur (Kaml et al., 1981). Since this fungus has a distinct identity, therefore, the present collection merits description as a new species *Alternaria banyan* Mall and Kumar sp. nov.. No species of *Drechslera* has been found to be reported on *Ficus* sp. *Drechslera yamadi* has been found to be reported on Mulberry Subramanium and Jain (1966) on same family from Meerut (U.P.). Since this fungus has also a distinct identity, therefore, the present collection merits description as a new species *Drechslera fici* Mall and Kumar sp. nov..

4.1. *Alternaria banyan* Mall and Kumar sp. nov.

Infection spot epiphyllous, thin, circular to sub-circular spreading on entire leaf surface, brown to black, 4-10 mm in diameter. Colonies epiphyllous, effuse covering the infection spot dark black. Mycelium of external and internal hyphae. Stromata present. Conidiophores arising in fascicles macronematous, mononematous, geniculate, straight or flexuous, cylindrical, unbranched up to 10 transverse septa, simple thick walled smooth, olive brown 40-110 x 4-7 μ m in diameter. Conidiogenous cells integrated, terminal to intercalary, sympodial, generally monotretic rarely polytretic, cicatrized, bearing thickened conidial scars. Conidia muriform, aeropteralogenous in chains on single, straight or slightly curved, up to 4 transverse septate with several longitudinal and oblique septa thick-walled, smooth, brown, base obclavate to rounded, hilum thickened 25-70 x 14-20 μ m in diameter; thick at broadest part, constricted at the septae, beak pale and septate or unseptate (Figure 1).

On living leaves of *Ficus benghalensis* Linn. (Moraceae), Gulra Rest House, Bhinga Forest Range, Shrawasti (U.P.) India, 01.04.2013, leg; Ajay Kumar, BRH-3652, AK-0152 (Isotype), HCIO- Holotype, Mycobank- MB - 805889

Maculae epiphyllae ciculares vel sub-circulares, extensae per totum foliae, brunnae vel atrae, usque at 4-10 μ m in diam. Coloniae epiphyllae, effuse, extidentes per totum contagionis maculae, atrae. Mycelium ex hyphis exterum et internum. Stromata evoluta. Conidiophora in fasciculo, macronemtosa, mononemtosa, geniculata, recta vel flexuosa, cylindria, non-ramosa, usque 10 transeverse septata, simplicia crassitunicata, glabra,

Species • RESEARCH • THREE NEW FOLIICOLOUS FUNGI

Table 2

The comparative account of *Asterina mysorensis* Hanf. and Thirum (1948) & *Asterina benghalensis* sp. nov.

| S. No. | Comparable Points | <i>Asterina mysorensis</i> Hanf. And Thirum (1948) | <i>Asterina benghalensis</i> sp. nov. |
|--------|-------------------------|---|--|
| 1. | Infection spots | Amphigenous 2-3 mm in diam. | Epiphyllous 4-8-10 mm in diam. |
| 2. | Mycelium, Hyphae, Cells | 6-8 μ m thick branched at 90° and 45° 20-30 μ m long. | 7-8 μ m thick, branched at 45° 25-30 μ m long. |
| 3. | Appresoria | Opposite or alternate 10-15 x 5-8 μ m | Opposite or alternate 10-12 x 5-7 μ m |
| 4. | Thyrothecia | Up to 130 μ m long. | 64-70-140 x 45-50-100 μ m |
| 5. | Asci | 8 spored 30 μ m in diam. | 4-6 spored 28 μ m in diam. |
| 6. | Spore | Single septate 26-32 x 14-16 μ m | Single septate 18 x 9 μ m |
| 7. | Remark | Collected from Karnataka | Collected from Shrawasti, (U.P.) |

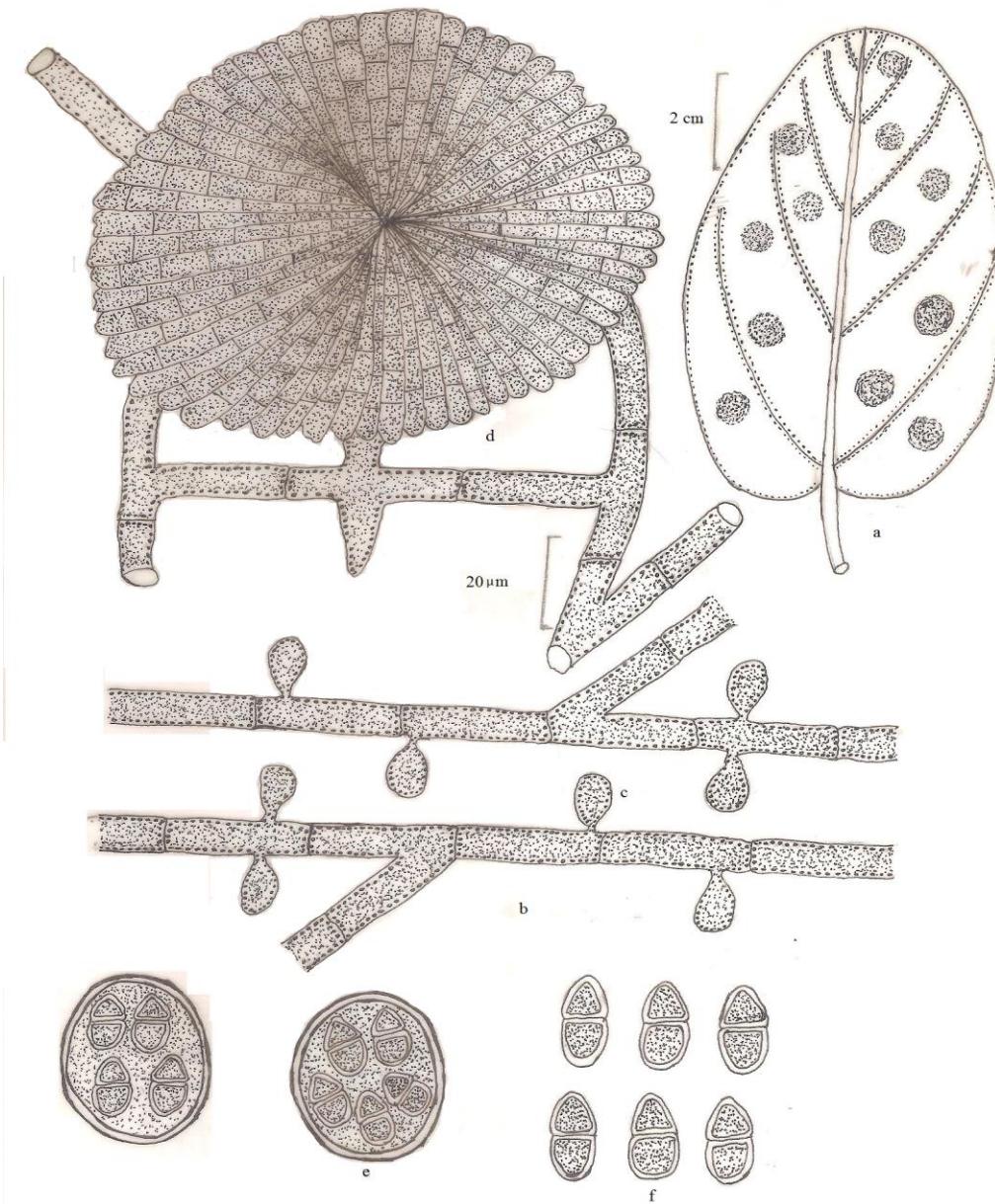


Figure 2

Asterina benghalensis Mall and Kumar sp. nov.; a) Infected leaf, b) Hyphae, c) Hyphopodia, d) Thyrothecium, e) Ascus with Ascospores, f) Ascospores

olivaceo-brunnea, 40-110 x 4-7 μ m in diam. Cellulae conidiogenae in Conidiophoribus incorporatae, terminalis vel intercalarius, sympodiales, frequenter monotreticatae, raro polytreticatae, cicatricatae, cicatrices conidiales incrassatae. Conidia muriformia, acropleurogenosa,

singulares vel catenatae, recta vel fere curvata, usque 4 transverses septatae vel vulgo plures longitudinalis vel vulgo plures longitudinalis vel obliquae septatae crassitunicata, glabra, brunnea, basin obclavata vel rotundata, hilo incrassato, 25-70 x 14-20 μ m in diam.

crassa ad evolutum partum, porce constricta ad septata, beak pallid et unseptata vel septata.

In foliis vivis *Ficus benghalensis* Linn. (Moraceae), Gulra Rest House, Bhinga Forest Range, Shrawasti (U.P.) India, 01.04.2013, leg; Ajay Kumar, BRH-3652, AK-0152 (Isotype), HCIO- Holotype, Mycobank- MB - 805889

A perusal of literature shows that only *Alternaria* sp. is reported on *Ficus benghalensis* from Khandala, Bombay (M.S.) by Uppal et al., 1935, but the description of the fungus is not given or available to us. *Alternaria tenuissima* is reported by Kamal et al., (1981) on *Artocarpus heterophyllus* Lamk. (Moraceae) from Gorakhpur (U.P.) India is the only species found to be comparable to the present collection. The comparative account is given in Table 1. Comparative account reveals that fasciculate, unbranched, geniculate conidiophores with thickened conidial scars and muriform conidia with broader obclavate to rounded base of the present collection are significantly different from *Alternaria tenuissima*. Therefore, it is worthwhile to propose the present collection as a new taxon of a species rank, to accommodate it. This specific epithet of *Alternaria banyan* is based on the host common name.

4.2. *Asterina benghalensis* Mall and Kumar sp. nov.

Colonies epiphyllous thin 4-8 up to 10 mm in diameter, dark brown to black, confluent, coalescing to cover almost entire leaf surface but the original shape remains intact; mycelium of dark brown hyphae 7-8 µm wide, the cells mostly 25-30 µm long branching opposite at 45°, closely reticulate; appresoria opposite or alternate, continuous, bluntly conoid with obtuse rounded apex 10-12 x 5-7 µm, thyrothecia densely scattered but aescrte black circular, convex up to 64-70-140 x 45-50-100 µm lower wall indistinct, upper wall of sub-opaque dark brown radiating hyphae; asci subglobose 4-6-8 spored sessile, about 28 µm in diameter; ascospore conglobate, oblong with rounded ends, subopaque dark brown single septate, constricted, 18 x 9 µm (Figure 2).

On living leaves of *Ficus benghalensis* Linn. (Moraceae), Gulra Rest House, Bhinga Forest Range, Shrawasti (U.P.) India, 01.04.2013, leg; Ajay Kumar, BRH-3652, AK-0152 (Isotype), HCIO- Holotype, Mycobank- MB - 805890

Coloniae ephyllae, effuse, densae, atrae, irregulars, partem majorem folii tegentes, demum coalescentes; mycelium superficial, pallid, brumeum, ex hyphis reticulates, crassitunicatis ad latera vel rare opposite ramosis, ecellules usque 25-30 x 7-8 µm. Compositum; appresoria unilateralia, appositie, unicellular 10-12 x 5-7 µm, capitata globosa vel hemiglobosa; thyrothecia dispersa vel aggregata, orbicularis, convexa medius, measure 64-70-140 x 45-50-100 µm ; asci orbiculares vel ovales, sessilis and 4-6-

8 sporae 28 µm in diam.; ascospores congestae elliptical oblong uniseptate, ad septatum constrictae, rotundate ad denum, sub caeruleus vel brumeolus flavae, glabro tunicate, 18 x 9 µm.

In foliis vivis *Ficus benghalensis* Linn. (Moraceae), Gulra Rest House, Bhinga Forest Range, Shrawasti (U.P.) India, 01.04.2013, leg; Ajay Kumar, BRH-3652, AK-0152 (Isotype), HCIO- Holotype, Mycobank- MB - 805890

Perusal of literature reveals that as yet no species of *Asterina* has been described on this specific host, the morphotaxonomic comparissin is done with *Asterina mysorensis* Hanf. And Thirum (1948) which is found to be comparable reported on *Ficus* sp. The comparative account is given in Table 2. The comparative account shows that the morpho taxonomic features of the present collection is distinctly different from those of *Asterina mysorensis* and this fungus is not cons specific with any species of *Asterina* hitherto described on Moraceae , its description as a new taxon of separate species rank is thought worthwhile. The specific epithet of *Asterina benghalensis* is based on the host name.

4.3. *Drechslera fici* Mall and Kumar sp. nov.

Infection spot epiphyllous thin dark brown to black, confluent coalescing to cover almost entire leaf surface with original shape intact; conidiophores simple, emerging solitary, dark brown to olivaceous, geniculate, bearing 3-6 conidia occasionally more, producing the first conidium at a distance of about 30 µm from the base 90-250 x 6-8 µm (av. 170 x 7 µm), 8-10 septate, septa 10-25 µm apart. Conidia straight or more often slightly curved to one side, tapering slightly towards both the abruptly rounded end, cylindrical, slightly broader near the middle, light fuligenoous when young later turning it into light to dark brown in color at maturity; 82 x 21 µm, 08 -11 septate, non constricted at the septum (Figure 3).

On living leaves of *Ficus benghalensis* Linn. (Moraceae), Gulra Rest House, Bhinga Forest Range, Shrawasti (U.P.) India, 01.04.2013, leg; Ajay Kumar, BRH-3652, AK-0152 (Isotype), HCIO- Holotype, Mycobank- MB - 805891

Infection spot ephyllae, effuse, dense, atrae, irregulars, patem majorem folii tegentes, demum coalescentes; Conidiophores simplices, solitaria, emergentia, atro brunnea vel olivacea, geniculata, 3-6 conidia instructor. Conidia suballantoidea med 170 x 7 µm, 8-10 septata, septa 10-25 µm se separata, conidia recta, persaepo lateraliter leviter curvata, ad ultramque rotundatum extremitatem abrupt angustata, cylindracea, ad medium leviter latores, primo pallid foliginen denum pallid atro-brunnea, 82 x 21 µm, 08-11 septate, adseptum non-constrictata.

Table 3

The comparative account of *Drechslera yamadi* Subramanium and Jain & *Drechslera fici* sp. nov.

| S. No. | Comparable Points | <i>Drechslera yamadi</i> Subramanium and Jain (1966) | <i>Drechslera fici</i> sp. nov. |
|--------|-------------------|--|---|
| 1. | Infection spot | Abaxial | Epiphyllous (Adaxial) |
| 2. | Conidiophores | Emerging singly or in tuft of 3-5, 3-8 septate, 20.8-78 x 5.2-9.1 μ m. | Emerging singly through stomata, 8-10 septate, 170 x 7 μ m Septa 10-25 μ m apart. |
| 3. | Conidia | 2-8 septate, 18.2-71.5 x 3.9-9.1 μ m. | 8-11 septate, 82 x 21 μ m. non constricted at the septum |

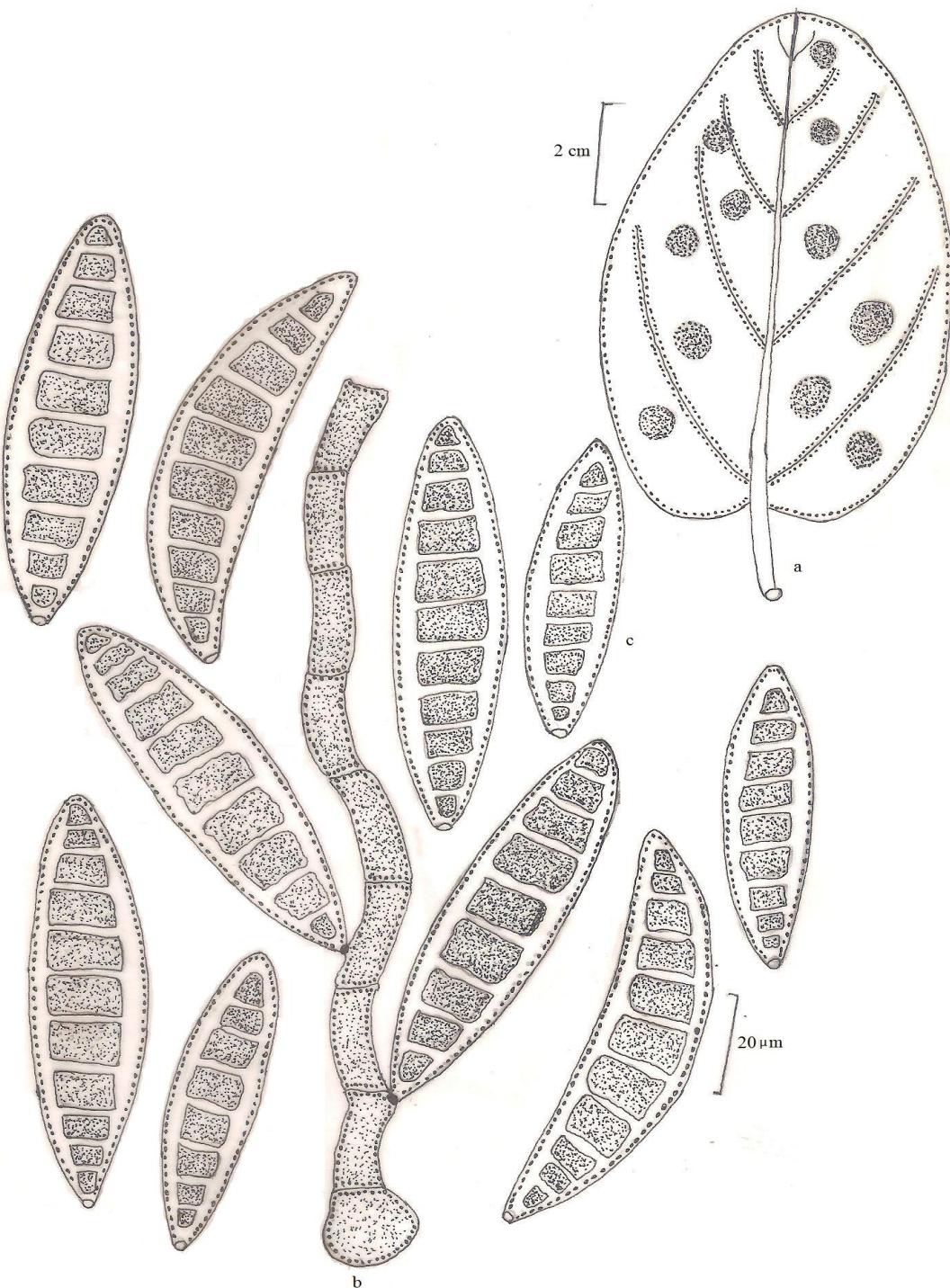


Figure 3

Alternaria banyan Mall and Kumar sp. nov.; a) Infected leaf, b) Conidiophores, c) Conidia

In foliis vivis *Ficus benghalensis* Linn. (Moraceae), Gulra Rest House, Bhinga Forest Range, Shrawasti (U.P.) India, 01.04.2013, leg; Ajay Kumar, BRH-3652, AK-0152 (Isotype), HCIO- Holotype, Mycobank- MB - 805891

Perusal of literature reveals that as yet no species of *Drechslera* has been described on this specific host, the morphotaxonomic comparissin is done with the morphotaxonomic comparissin is done with *Drechslera yamadi* Subramanium and Jain 1966 which is found to be comparable reported on Mulberry (Moraceae) from Meerut (U.P.). The comparative account is given in Table 3. The above comparative account shows that the morpho taxonomic features of the present collection is distinctly different from those of *Drechslera yamadi* and this fungus is not cons specific with any species of *Drechslera* hitherto described on Moraceae , its description as a new taxon of separate species rank is thought worthwhile. The specific epithet of *Drechslera fici* is based on the host name.

The review of Literature Bilgrami et al., (1979), (1981), (1991); Ellis (1971), (1976); Ellis and Ellis, (1997); Hansford et al., (1948); Hosagaudar et al.,

(1996), (2006), (2012); Jamaluddin et al., (2004); Kamal et al., (1981); Mukerji et al., (1974); Sarbhoy et al., (1986), (1996); Subramanium and Jain (1966); Verma et al., (2008); Uppal et al., (1935) reveals that there is no record of *Alternaria banyan*, *Asterina benghalensis* and *Drechslera fici*. Therefore, it is described and illustrated as a new species to accommodate them.

5. CONCLUSION

The region of Shrawasti, U. P. is rich in phytodiversity in general as well as the diversity of fungal organisms inhabiting plant leaves in particular and it provides great scope for study of foliicolous fungi. Correct identity of a fungus absolutely free from ambiguities is vital for its employment in applied disciplines in general and it is more so for plant pathology where precision of details about the biology of the pathogen is primarily conditioned by its identity. In fact, without being equipped for ascertaining the correct identity of a fungal pathogen all studies concerning its phyto pathological aspects would simply be misleading. However the end is still not insight and further investigation is warranted.

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